# EVALUATION OF BARRIER TREATMENTS ON NATIVE VEGETATION IN A SOUTHERN CALIFORNIA DESERT HABITAT

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ABSTRACT. Treating perimeters with residual insecticides for protection from mosquito vectors has shown promise. These barrier treatments are typically evaluated in temperate or tropical areas using abundant vegetation as a substrate. However, there is an emerging interest to develop this technology to protect deployed US troops in extreme desert environments with sparse vegetation. We used a remote desert area in the Coachella Valley, California, to 1) evaluate bifenthrin barrier treatments on native xeric vegetation and 2) compare treatments applied with electrostatic and conventional spray technologies. Through a combination of laboratory bioassays on treated and control vegetation sampled at specific intervals over 63 days, synchronized with field surveillance of mosquitoes, we measured the temporal pattern of bioactivity of bifenthrin barriers under natural hot, dry, and dusty desert conditions. Regardless of spray technology, mosquito catch in treated plots was about 80% lower than the catch in control plots 1 day after treatment. This reduction in mosquito numbers in treated plots declined each week after treatment but remained at about 40% lower than control plots after 28 days. Field data were corroborated by results from bioassays that showed significantly higher mosquito mortality on treated vegetation over controls out to 28 days postspray. We concluded that barrier treatments in desert environments, when implemented as part of a suite of integrated control measures, may offer a significant level of protection from mosquitoes for deployed troops. Given the comparable performance of the tested spray technologies, we discuss considerations for choosing a barrier treatment sprayer for military scenarios.

**KEY WORDS** Electrostatic sprayer, residual pesticide, mosquito-borne disease, bifenthrin, Deployed Warfighter Protection Program (DWFP)

#### INTRODUCTION

Treating perimeters of vegetation or an artificial substrate with residual insecticides to provide protection from mosquito and sand fly disease vectors, as well as nuisance arthropods, has shown promise, with an intermittent record of research dating back over 60 years (Madden et al. 1947, Quarterman et al. 1955, Pant and Joshy 1969, Eshghy and Nushin 1978, Helson and Surgeoner 1983, Robert and Perich 1995, Orshan et al. 2006) and a recent surge in popularity (Royal 2004, Hubbard et al. 2005, Cilek and Hallmon 2006, Frances 2007, Trout et al. 2007, Cilek 2008, Faroog et al. 2008). Examples of treated perimeter vegetation include jungle, forest, or hedges surrounding a house, village, or park (Anderson et al. 1991, Perich et al. 1993) or even open grasslands (Kettle 1949). Examples of treated artificial perimeter substrates include interior and exterior walls (Huehne 1971, Lee

et al. 1997), suspended or spread sheets (Elnaiem et al. 1999, Graham et al. 2002), bed nets (Hill et

focused on protecting permanent human or livestock dwellings against common pest or vector insects in temperate or tropical regions. Current US military operations in hot, dry, and dusty environments containing persistent threats from disease-transmitting arthropods have brought about a growing interest in transferring barrier treatment technology to deployed military personnel stationed in temporary shelters in harsh, barren desert terrain (Linthicum et al. 2007, Cope et al. 2008, Dalton 2008). A key strategic advantage of developing a barrier treatment effective in harsh desert environments is that the residual chemical barrier could provide rapid long-term protection in nearly any location having regular or irregular perimeters of vegetation. Fortunately, a range of application technol-

al. 2006), and livestock fencing (Bauer et al. 2006a, 2006b). A range of insecticides has been used, from DDT in early studies (Lindquist and McDuffie 1945, Trapido 1947, Ludvik 1950, Nair 1951) to various pyrethroids and other toxicants in more recent work (Sathantriphop 2006, Xue 2008). Experimental barrier treatments have mainly targeted mosquitoes, especially vectors of malaria (Yadav 2003, Diabate et al. 2006) but have also been assayed against sand flies (Jacusiel 1947, Perich et al. 1995, Kelly et al. 1997) and biting midges (Kettle 1949, Royal 2004).

In large part, barrier treatment studies have

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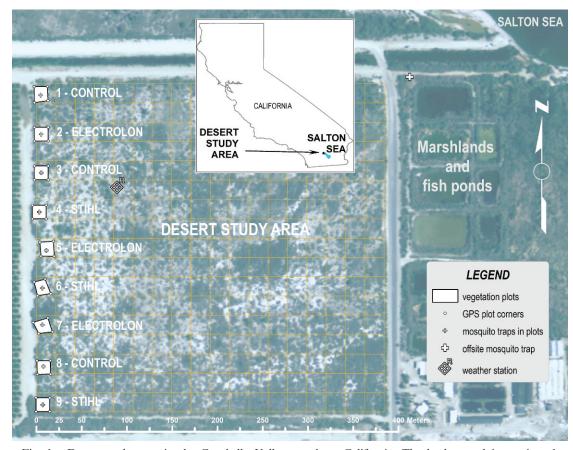


Fig. 1. Desert study area in the Coachella Valley, southern California. The background image is a 1-m resolution natural color Digital Orthophoto Quadrangle (NAD 1983, UTM Zone 11N) available from the USGS at http://seamless.usgs.gov. The shore of the Salton Sea is visible in the northeast corner of the image, with the fish hatchery ponds and marshlands to the east of the study area. White squares on the image show the shape and location of the 9 study plots, and labels indicate Electrolon or Stihl treatment or control. Gray cross symbols within squares show locations of mosquito traps; the single white cross symbol at the northwest corner of the fish ponds shows the offsite control mosquito trap. The "R" on the weather station symbol marks that a weather data recorder was present.

ogies exists for carrying out barrier treatments in the deployed military environment. In this study, we used a desert field site to investigate differences in performance between 2 spraying systems, a standard mist blower and an electrostatic mist blower, and concurrently evaluate the feasibility and efficacy of barrier treatments in a desert environment

# METHODS AND MATERIALS Study site

We used a large desert area in the Coachella Valley, California, to evaluate spray technology and barrier treatments of bifenthrin on native xeric vegetation under hot, dry, and dusty field conditions (Fig. 1). This natural study site is situated just west of a cluster of active and abandoned fish ponds and marshy areas in a

region gridded with active canals and 0.5 km from the northwest shore of the Salton Sea  $(33.46^{\circ}N, 116.06^{\circ}W; -64 \text{ m})$ . The effluent from commercial fish ponds and seasonal flooding of marshy areas along the Salton Sea create a large and highly productive habitat for the development of wild *Culex tarsalis* Coquillett. As the Salton Sea level rises annually from February through May, Cx. tarsalis development is triggered (Reisen et al. 1995). Owing to the March 2008 spring timing of this study we expected abundant wild seasonal production of adult mosquitoes and a high biting pressure to be present. Salton Sea surface elevation data, an index of water level, from the Coachella Valley Water District (CVWD, PO Box 1058, Coachella, CA 92236) later confirmed these expectations (Fig. 2). We delineated nine 15.25 m  $\times$  15.25 m vegetation study plots spaced approximately

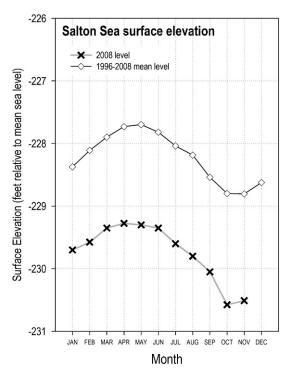


Fig. 2. Monthly mean Salton Sea surface elevation, an indication of water level, in feet relative to mean sea level. The annual rise in water level from February to May triggers the development of *Culex tarsalis* in shore habitat. The overall low water level in 2008 compared to the long-term mean reflects an ongoing and gradual annual decline over the last decade. Nevertheless, the trend of increasing water level in spring 2008 indicates the presence of natural breeding habitat for wild *Cx. tarsalis* throughout the 28-day population sampling period in the desert study area.

25 m apart, situated north to south along the western edge of the field to take advantage of a dirt access track (Fig. 1). We first mapped study plots using a 1-m resolution 3-band (RGB) natural color Digital Orthophoto Quadrangle (DOQ; available from the USGS, http://seamless. usgs.gov) in an ArcGIS 9.2 (Environmental Systems Research Institute, Redlands, CA) geographic information system (GIS), extracted coordinates of corner and center points using

the GIS, and finally marked the plots on the ground using a geographic positioning system (GPS) and pin flags. We surveyed and recorded points in the field using a GeoXT (Trimble, Sunnyvale, CA) handheld GPS, which operated at approximately 3 m precision (uncorrected) and was set at the NAD 1983 datum for spatial reference to UTM Zone 11N to match the USGS DOQ. We used a small tracked vehicle to flatten a swath through vegetation around the marked plots to permit uniform access for the sprayers around the plot boundaries. The dominant plant species at the study site included Tamarix chinensis Lourteig (salt cedar), Pluchea sericea (Nuttall) Coville (arrow weed), Atriplex canescens (Pursh) Nuttall (salt bush), and Salicornia spp. (pickle weed).

#### **Barrier treatments**

The barrier treatments consisted of applications of bifenthrin using 2 sprayer models, the Stihl® SR-420 backpack sprayer and the Electrolon® BP-2.5 electrostatic sprayer, and untreated controls. We partitioned the vegetation plots with a stratified random design, accounting for sparse, medium, and dense desert vegetation, for 3 treatment replicates with each sprayer and 3 replicates of untreated controls. We performed treatments with 2 different application technologies to evaluate whether they would influence the efficacy of the barriers. We applied Talstar® bifenthrin (FMC Corp., Philadelphia, PA) in water at the label rate of 1.0 ounce per 1,000 ft<sup>2</sup>, and walking speed was adjusted to account for variation in flow rate between the 2 sprayers to finish with near-identical volume of active ingredient on all treated plots (Table 1). Spray teams applied bifenthrin using the GPS-surveyed pin flags as guides and endeavored to cover as many individual plants as possible along the plot boundaries within the allotted spray time. A portable Integrated Sensor Suite weather station with a Vantage Pro2 data recorder (Davis Instruments, Hayward, CA) was erected at 2 m on a pole within the field (Fig. 1). Figure 3 shows weather patterns for the duration of the study from this weather station. We measured the

Table 1. Details of spray activity.

Treatment/equipment	Control (plots 1, 3, 8)	Electrolon BP-2.5 (plots 2, 5, 7)	Stihl SR 420 (plots 4, 6, 9)
Walking speed	_	0.10 mph	1.39 mph
Calibrated flow rates	_	6.8 oz/min	93.8 oz/min (setting 2)
Weather conditions at time of	_	~75°F	~75°F
spray <sup>1</sup>		~25% RH	~25% RH
		4.0-6.0 mph wind speed	4.0-6.0 mph wind speed

<sup>&</sup>lt;sup>1</sup> March 19, 2008 (1000–1100 h). Weather data from Thermal Airport, Thermal, CA (available from the Weather Underground, http://www.wunderground.com), approximately 17 km northwest of the study area, are presented because the onsite weather station recorder malfunctioned on this day.

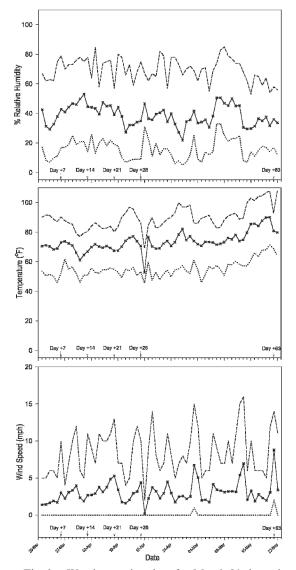


Fig. 3. Weather station data for March 21 through May 22, 2008. Sample days relative to day of treatment are marked above the x axis. No precipitation was detected in this sample period. Data are missing from Day -1 (18 March), Day 0 (19 March), and Day +1 (20 March) because of a malfunction of the weather data recorder.

temporal pattern of bioactivity of the bifenthrin barrier treatments under desert conditions using a combination of bioassays on foliage sampled at specific time intervals from these plots over 63 days, synchronized with field surveillance of mosquitoes within treatment and control plot areas.

# **Sprayers**

The Electrolon BP-2.5TM (Electrostatic Spraying Systems, Watkinsville, GA) is a backpack

electrostatic mist blower that requires pressurized air from an external source with a minimum supply of 8.1 cfm at 60 psi. We used a Terminator diesel compressor (Adapco, Sanford, FL) to supply air to the Electrolon. The sprayer uses an air-assistance induction charge nozzle. The liquid to be applied is fed from the tank by gravity and siphoned to the handgun by movement of the pressurized air forced out of the nozzle. The force of the pressurized air shatters the liquid at the nozzle to form the spray mist, and the spray droplets are negatively charged by two 9-V rechargeable batteries. The Electrolon BP-2.5 sprayer has a net weight of 9 lb, a pesticide tank capacity of 4 gallons, and a flow rate of approximately 6.8 oz/min.

The Stihl SR 420 (Andreas Stihl, Waiblingen, Germany) is a backpack mist blower powered by a 3.4-hp single-cylinder 2-cycle Stihl engine, with the capability to produce an air flow rate of 625 cfm and an air velocity of 180 mph. The sprayer uses an air-shear atomization head with screens to alter the spray release pattern. The flow rate can be set from a control knob placed near the head that has 6 metering nozzle settings ranging from 4.7 to 100 oz/min. Setting 2 was used for this study, which produced 93.8 oz/min. The Stihl SR 420 has a net weight of 24 lb, a pesticide tank capacity of 3.7 gallons, and the bystander noise level is 75 dBA.

# Vegetation samples

We sampled vegetation from the nine plots the day before (Day -1), the day of (Day 0), and 1, 7, 14, 21, 28, and 63 days after the spray and carried out bioassays in the laboratories at the Coachella Valley Mosquito and Vector Control District (CVMVCD). Wearing nitrile gloves we gathered vegetation samples by plot into separate, labeled sample bags and changed gloves and cleaned all cutting instruments with 90% ethanol between plots to minimize cross-contamination of samples. Sample bags were constructed of USDA IR-4-approved inert material developed for storing samples of vegetation without contamination for chemical analysis (Hubco, Hutchinson, KS). We collected 20  $\sim$ 12 cm sprigs of vegetation cut in a nonuniform fashion from all 4 sides of each plot, which would provide enough material for 10 bioassays for each plot. We stowed all sample bags in insulated coolers on retrieval from the field and quickly transferred them to an ultralow freezer in the laboratory to minimize bifenthrin degradation after sampling.

# Mosquito collections

For mosquito population sampling in the field we set modified Encephalitis Virus Surveillance mosquito traps (Rohe and Fall 1979) baited with dry ice (CO<sub>2</sub>) without light at the centroid of each plot on a permanent stanchion and ran them overnight (ca. 1700–0800 h) each day vegetation was sampled. One exception was that no mosquito trapping was performed at Day +63 because of extremely windy conditions (Fig. 3). A fourth offsite control trap was set ca. 1,400 ft away on the east side of the field close to immature mosquito development sites (Fig. 1). Traps were collected the next morning and mosquitoes transported to the lab for identification and counting, and then archived in labeled Petri dishes in a refrigerator.

#### Bioassay

The bioassay setup consisted of placing a single cut sprig into a labeled 30 mm × 140 mm glass culture tube with ten 3- to 5-day-old coldanaesthetized female Cx. tarsalis. Culex tarsalis colony mosquitoes were reared at the CVMVCD at 82°F and 49% RH and given a 10% sugar water solution for nourishment. The Cx. tarsalis mosquitoes in the CVMVCD colony originated from the "BSF" colony established in 1952 by W. C. Reeves from mosquitoes collected around Bakersfield (Kern County), CA. The BSF colony population is considered to not have ever been exposed to chemicals currently used in mosquito control. We sealed tubes with white polyester no-See-Um netting (Skeeta, Bradenton, FL) held in place with 2 silicone O-rings and stored them horizontally in metal racks shelved in the mosquito-rearing room, which was maintained at 82°F and 49% relative humidity throughout the experiment (Fig. 4). The setup was designed to exploit the xeric nature of the vegetation, in that the tough, woody stems and small rigid leaves would stand up independently in the tubes and provide an attractive, natural resting site for mosquitoes. Preliminary observations of the bioassay confirmed that female Cx. tarsalis mosquitoes used in the experiment overwhelmingly preferred to rest on the vegetation over the glass or netting cap, ensuring contact with plant surfaces that might contain bifenthrin. We carried out 10 bioassay replicates per plot for each sample day and recorded mortality of female mosquitoes in the tubes at 6, 24, and 48 h.

#### RESULTS

The results of the bioassay (Fig. 5) provide a detailed temporal record of the bioactivity of the bifenthrin barrier treatment as measured by mortality of female *Cx. tarsalis* mosquitoes exposed in the lab to treated vegetation collected from the field. The 3 graphs show data for the same mosquitoes inspected after 6, 24, and 48 h exposure to the treated vegetation. In all graphs the lowest curve, dashed line, shows mosquito

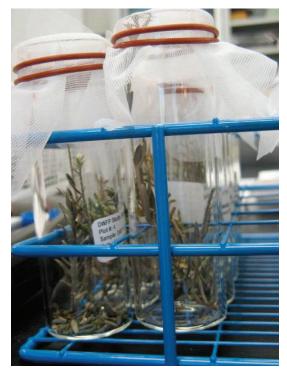


Fig. 4. Bioassay apparatus consisting of labeled 30 × 140 mm glass culture tubes in a wire rack, capped with polyester no-see-um netting held fast with silicone O-rings. Vegetation samples naturally stand upright in tubes providing an attractive resting site for female mosquitoes. Mortality is easily tallied with excellent visibility into all parts of the tube.

mortality data on the untreated control vegetation. Overall, mortality in bioassays on vegetation from both spray technologies was significantly higher than mortality of female mosquitoes on untreated vegetation out to 28 days posttreatment (P < 0.001, Kruskal-Wallis one-way analysis ofvariance on ranks; P < 0.05, Tukey multiple comparison test). Error bars on the graphs show standard errors of means, and nonoverlapping error bars correspond to a significant difference between treatments supported at P < 0.05 (Tukey multiple comparison test). These graphs show that with some variation between them, not consistent week to week, the electrostatic and standard mist blowers appear to perform equally well in the tested environment. For both spray technologies, although mortality at 24 h dips only below 50% after about 14 days posttreatment, 48 h mortality remains at 50% or above out to 28 days posttreatment. In samples from Day +63, mortality at 48 h exposure on treated vegetation converges with mortality on untreated vegetation for both spray technologies.

The results of field mosquito population surveillance performed from Day -1 to Day +28 (Fig. 6) largely corroborate the findings of

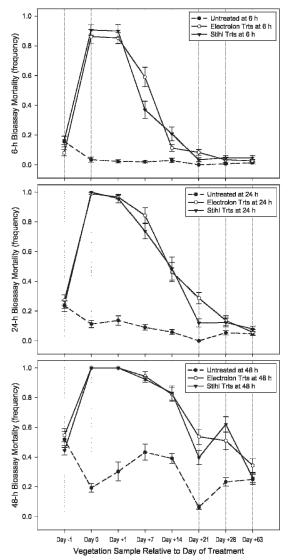


Fig. 5. Results tracking mortality of female Cx. tarsalis at 6, 12, and 24 h exposure in bioassays on vegetation sampled March–April 2008 from plots in the desert study area. Error bars represent standard errors of means, and nonoverlapping error bars show significant difference supported at P < 0.05 (Tukey multiple comparison test). The bottom curves in each graph show mortality on untreated control vegetation. Day 0 = day of barrier treatment.

the bioassay. For example, bioassay mortality in the lab at 48 h for samples from Day +28 was about 40% higher in treatments compared to controls, and field trapping showed a reduction of about 40% at Day +28 in treated plots as compared to untreated control plots. Biting pressure as indicated by trap counts of female mosquitoes was high throughout the study at all trap locations (Table 2), providing evidence that

low trap numbers in treated plots result from the chemical barrier and not local population fluctuations. The histograms in Figure 6 point to the Electrolon treatment being marginally, although not significantly, more effective than the Stihl treatment from Day 0 to Day +28. The negative values for Day -1, the day before treatment, and for Day +21 in the Stihl plots shows that those plots happened to have more trapped mosquitoes than the control plots on those days. The histogram for the Stihl plots in Figure 6 does not include data from plot 9 because of unusually high trap counts that were clearly outliers compared to all other plots. Plot 9 was thicker with vegetation than any other plot and was adjacent to a very heavily vegetated area to the south of the desert field, possibly providing relatively cool, shady, and humid refugia for adult mosquitoes. Although mosquito numbers were somewhat reduced in Plot 9 after treatment, the population pressure was beyond the capacity of the treatment to control effectively.

#### DISCUSSION

The results of the bioassay suggest that barrier treatments are not just effective in the desert environment, but are effective and enjoy similar longevity regardless of application technology. This result was particularly interesting because the performance of the Stihl sprayer surpassed the performance of the Electrolon sprayer during other studies in humid environments in Arkansas (Kline et al. 2007) and Florida (Farooq et al. 2008). The performance of the Electrolon in these earlier studies as measured by bioassays was comparable to the current study, which indicates the Stihl may not perform equally in temperate and desert environments.

Given that the amount of bifenthrin sprayed by either technology on sampled foliage may vary within a plot on a given sample day, not only from inevitable unevenness in spraying and growth of new foliage but also from variation in sampling, we expected some variance in mortality from treated plots. This expectation is reflected by the taller bars of standard errors of the means, and thus greater variance, for bioassays on treated vegetation samples compared to bioassays on untreated vegetation at 6 and 24 h in Figure 5. Despite the variation, mortality in bioassays on treated vegetation was significantly higher out to 28 days posttreatment for both application technologies when compared to the controls. Interestingly, mortality of mosquitoes placed on vegetation treated with the Electrolon sprayer was still marginally significantly greater than mortality on untreated vegetation out to Day +63 at 48 h exposure.

One worthy target of improvement would be to develop the barrier treatment system to produce

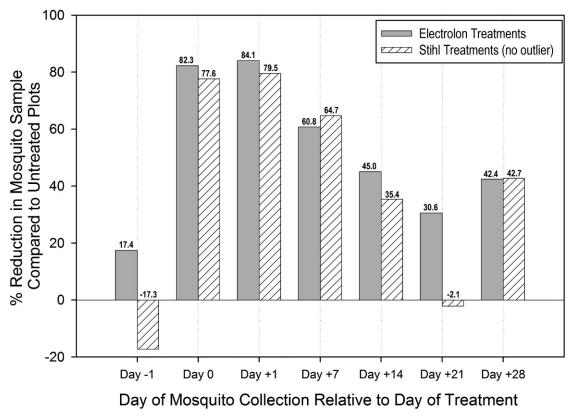


Fig. 6. Results for mosquito surveillance conducted March–April 2008 in the desert study area. Numbers above bars are the percent reduction in mosquito samples trapped in treated plots as compared to those trapped in control plots. For example, for Day +1 in plots treated with the Electrolon sprayer, we trapped over 84% fewer mosquitoes than in control plots in Day +1. See Table 2 for trap counts throughout the study. Excessive numbers of mosquitoes trapped at the heavily vegetated Stihl treatment plot 9 created an outlier that was excluded to produce this histogram. Day 0 = day of barrier treatment.

treatment bioassay results in the quick-kill 6 h observations that are currently seen here at the long-term kill 48 h observations. However, observations in the lab during bioassays suggest that mosquitoes contacting treated foliage, if not rapidly killed, display a behavior of disorientation and erratic movement, and are observed to lose legs, before eventual death. These observations suggest females contacting treated surfaces would quickly be removed from the host-seeking cohort in the field. This was consistent with the observed absence of post-collection mortality in trapped mosquitoes. Thus, effective mortality, and therefore level of protection to people within the treated perimeter, may be higher than that inferred from absolute mortality in this study. In future studies it would be informative to examine host-seeking behavior of a variety of species and determine whether females are compelled to rest on substrates, therefore becoming available as targets of the barrier treatment (Perich et al. 1993), during movement toward a detected trap or human/animal host. To some extent this could be measured indirectly by comparing species diversity in trap samples before and after treatments, and species diversity in trap samples from control versus treated plots. However, this was not possible here because the collections in this study were >99% Cx. tarsalis.

The results of field mosquito population surveillance during the study (Fig. 6) are also very encouraging for the question of efficacy of barrier treatments in the desert environment and show that, concordant with the bioassay results, both application technologies have merit in the desert environment. Salton Sea water level throughout the mosquito surveillance period (Fig. 2) indicates that natural breeding sites for Cx. tarsalis were still available and thus do not account for the reductions in population samples. Interestingly, the atypical low reduction at Day +21 in both Electrolon and Stihl plots may indicate the arrival of a new cohort of mosquitoes from nearby breeding areas. Weekly CVWD Salton Sea water level measurements continued to show an upward trend through April (data not shown), and it is possible a population surge of Cx. tarsalis took place. If it is the case that a new

Culex tarsalis collected in mosquito traps in the 9 study plots. Counts are tallied by type of barrier treatment and day of sample relative to barrier treatment. No trapping was performed at Day +63 because of conditions of extreme wind

				Mean (min-max; N)	(Z		
Plot/sample	Day -1	Day 0	Day +1	Day +7	Day +14	Day +21	Day +28
Control Electrolon Stihl Stihl (No plot 9)	Control 363.7 (234-586; 3) Electrolon 300.3 (113-514; 3) Stihl 665.3 (346-1,143; 3) Stihl (No plot 9) 426.5 (346-507; 2)	203.5 (32–584; 4) 36 (11–62; 3) 90 (40–179; 3) 45.5 (40–51; 2)	205 (102–453; 4 32.7 (5–66; 3) 128.3 (35–301; 3) 42 (35–49; 2)	5 (32–584; 4) 205 (102–453; 4) 650.8 (199–1,512; 4) 66 (11–62; 3) 32.7 (5–66; 3) 255.3 (99–458; 3) 80 (40–179; 3) 128.3 (35–301; 3) 630.3 (180–1,432; 3) 55 (40–51; 2) 42 (35–49; 2) 229.5 (180–279; 2)	616 (244–1,128; 4) 338.7 (160–572; 3) 662.7 (372–1,192; 3) 398 (372–424; 2)	503.3 (145–1,000; 4) 349.3 (116–736; 3) 886.7 (388–1,632; 3) 514 (388–640; 2)	363.7 (234–586; 3) 203.5 (32–584; 4) 205 (102–453; 4) 650.8 (199–1,512; 4) 616 (244–1,128; 4) 503.3 (145–1,000; 4) 2,060 (1,032–3,968; 4) 300.3 (113–514; 3) 36 (11–62; 3) 32.7 (5–66; 3) 255.3 (99–458; 3) 33.7 (160–572; 3) 349.3 (116–736; 3) 1,186.7 (792–1,392; 3) 365.3 (346–1,143; 3) 90 (40–179; 3) 128.3 (35–301; 3) 630.3 (180–1,432; 3) 662.7 (372–1,192; 3) 886.7 (388–1,632; 3) 1,690.7 (1,072–2,712; 3) 426.5 (346–507; 2) 45.5 (40–51; 2) 42 (35–49; 2) 229.5 (180–279; 2) 398 (372–424; 2) 514 (388–640; 2) 1,180 (1,072–1,288; 2)
<sup>1</sup> Day $0 = \text{day of}$	Day $0 = \text{day of barrier treatment.}$						

cohort arrived in the area on Day +21, it is reassuring to observe the rebound in level of control at the treated plots a week later at Day +28. On the other hand, bioassay data for samples from Day +21 at 48 h show a reduction in mortality in both treated and control vegetation that is anomalously low compared to the trend at 48 h from Day +14 and Day +28 samples. This anomalous reduction could be due to the vagaries of sampling. However, a change in a weather parameter or a change in plant physiology between Day +14 and Day +21 could have altered the bioavailability of bifenthrin to the target mosquitoes that was somehow preserved in the frozen vegetation sample but permitted to change in the vegetation in the field. Unfortunately the weather record in Figure 3 does not reveal any obvious changes that may have affected the bifenthrin. Vigilance for similar phenomena in future experiments should be maintained.

One critical issue in evaluating barrier treatments is whether females arrive in traps despite having contacted treated surfaces, in which case we must assume they would have attempted to bite a person within the protected area. Although this study was not designed to measure whether trapped females had been exposed to bifenthrin, we estimated mortality in trapped mosquitoes from Day 0 and Day +1 at 12, 24, and 48 h before removing them for counting and archiving. We kept the containers containing the trapped mosquitoes in the warm, humid mosquito-rearing room and supplied cotton balls soaked in 10% sugar solution, but did not observe particularly excessive mortality in trapped females from treated plots versus control plots. We hypothesized that females reaching traps in treated plots had either not made sufficient contact with treated vegetation to obtain a lethal dose of bifenthrin or were resistant to the chemical. Information on resistance to pyrethroids in wild Cx. tarsalis is sparse, but data from Strong et al. (2008) suggest that permethrin is still effective against populations of Cx. tarsalis in northern

In any case, the fact that female host-seeking mosquitoes still penetrate treated perimeters, although reduced in numbers, highlights the fact that as with many mosquito control measures, we stress that barrier treatment technology should be implemented as part of a suite of integrated control measures and not solely relied upon. Companion measures should include ULV or thermal fog treatment, personal protection with products containing DEET or other Environmental Protection Agency–approved compounds and clothing treated or impregnated with permethrin, removal trapping within the perimeter, source reduction outside and within the perimeter, and barrier treatment of artificial surfaces

within the perimeter. On the other hand, even in the absence of an integrated program of control, the results of the field mosquito population surveillance during the study allow us to hypothesize that a 40-80% reduction in mosquitoes crossing the treated barrier could translate into a 40–80% reduction in risk of exposure to mosquito-borne diseases for people within the protected area, compared to people situated nearby in untreated areas. Our study demonstrates that barrier treatments on vegetation in desert environments show great promise and should be investigated further. The Department of Defense does not specifically define standards for an effective treated barrier, but based on our results we may arbitrarily define "effective" as statistically significant higher mortality in treatments than controls in bioassays at 24 h, and ≥50% reduction in mosquito counts in traps placed in the field for 7–14 days postspray in treated plots as compared to control plots on the same day. Future work should aim to raise the bar on this initial standard and should include trials with a variety of perimeter sizes and multiple concentric perimeters to develop guidelines for optimal configurations.

In this study we have made a step toward evaluating barrier spray equipment that could be used in force health protection scenarios during troop deployment in desert environments. Through bioassays on treated vegetation and field sampling of mosquitoes, the efficacy of both the standard and electrostatic spray technologies were found to be comparable in the tested desert environment. However, there are important organic differences with military tactical significance between the technologies that should be considered apart from performance. The Electrolon, despite its small size, requires an external pressurized air source, and the operator must be tethered to the source by a heavy air hose. Air compressors of sufficient power to drive the Electrolon are noisy and must be vehicle mounted, and the air hose may limit movement of the operator through the environment. As consequence of a low flow rate (Table 1) the Electrolon has a much lower work rate than the Stihl, and the operator must move slowly and exercise more care in aiming the spraying wand at vegetation, and thus spend longer moving through the environment to perform the barrier treatment. However, the electrostatic spraying unit itself is light and operates with a hissing sound that is less noisy than the conventional sprayer, although the air compressor is loud. The Stihl is heavier and is loud but produces a higher flow rate (Table 1), which means that the operator may move much more quickly through the area to be treated and use less care in aiming the spraying wand at vegetation. Another consideration is that the Stihl does not appear to perform equally in temperate

and desert environments; however, the Stihl remained at least as effective as the Electrolon in both environments. In its current configuration, and given that its performance did not greatly surpass that of the Stihl, the Electrolon may not be the first choice for desert barrier treatments, especially if large areas are to be treated in a limited time in a tactical environment.

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